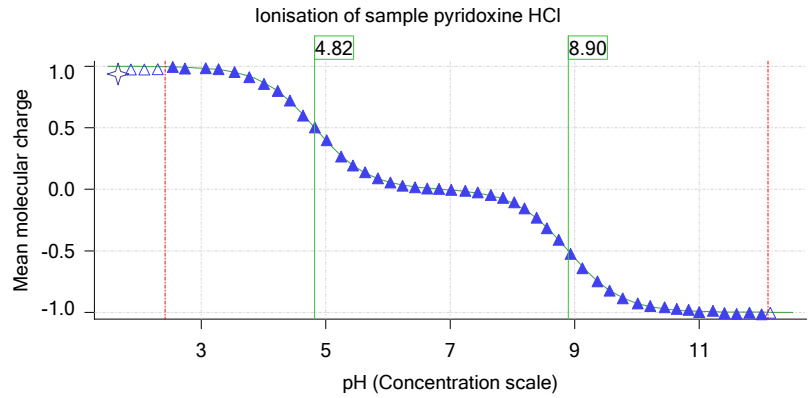
Sample name: **pyridoxine HCl**
Assay name: **pH-metric pKa**
Assay ID: **17I-11015**
Filename: **C:\Sirius_T3\Mehtap\20170911_test_comp_pKa\17I-11015_pyridoxine HCl_pH-metric pKa.t3r**

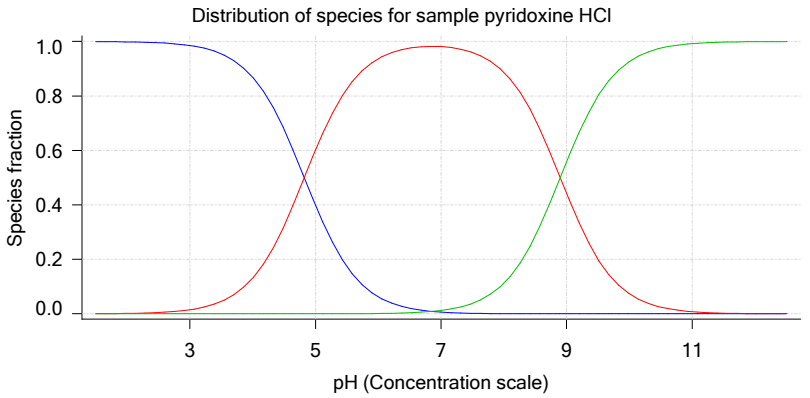
Experiment start time: **9/11/2017 4:00:50 PM**
Analyst: **Dorothy Levorse**
Instrument ID: **T311053**

## Sample

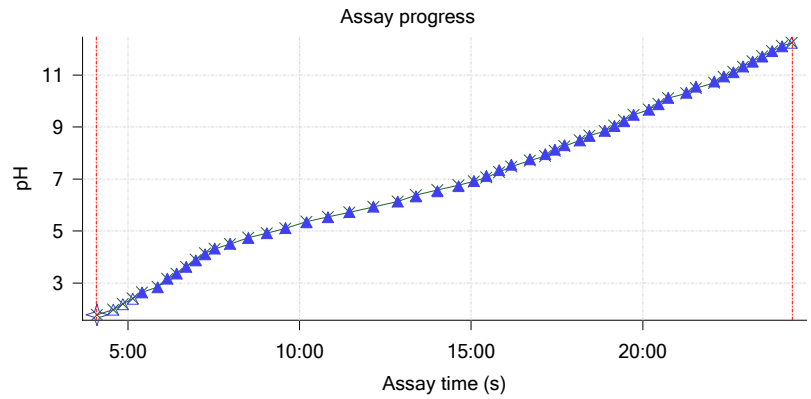
| | |
|---|---|
| pyridoxine HCl concentration factor | 0.964 |
| pyridoxine HCl stoichiometry | 1.000 |
| Chloride stoichiometry | 1.000 |
| Base pKa 1 | 4.82 |
| Acid pKa 2 | 8.90 |

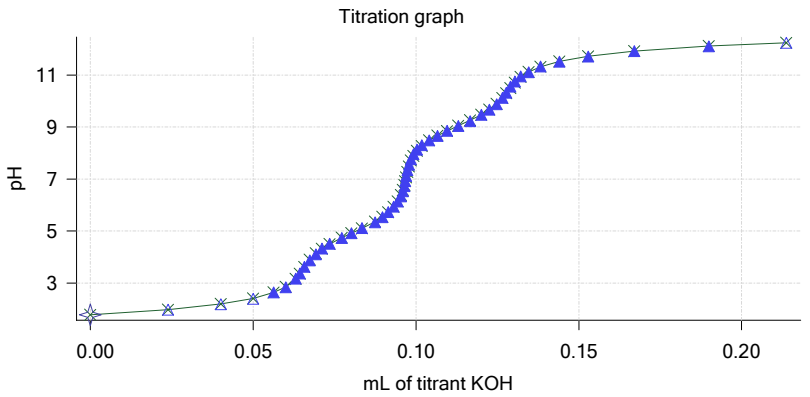
## Sample graphs

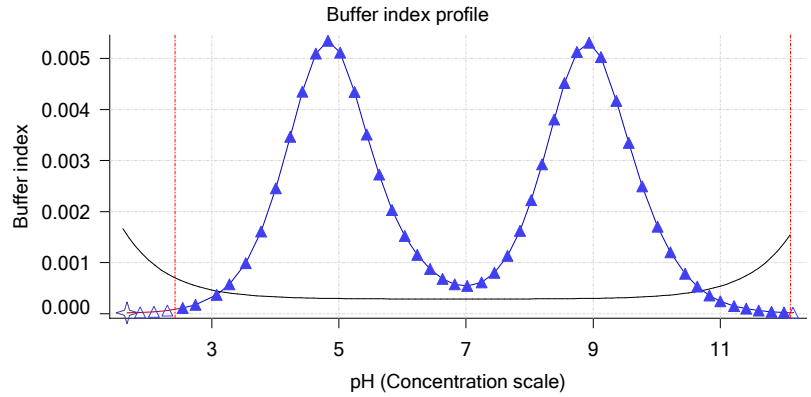


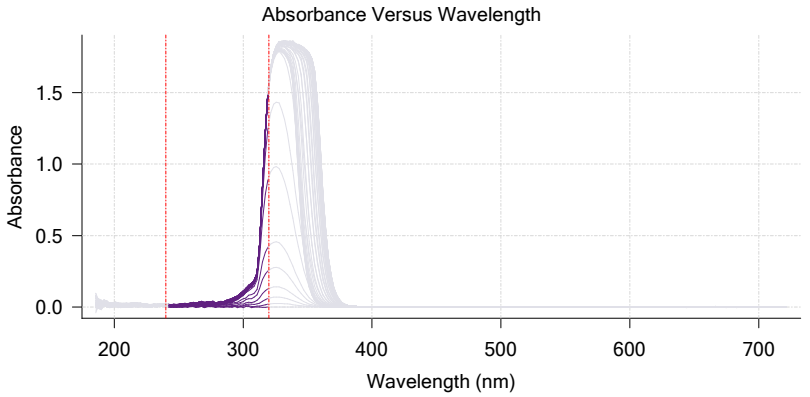




## Other graphs



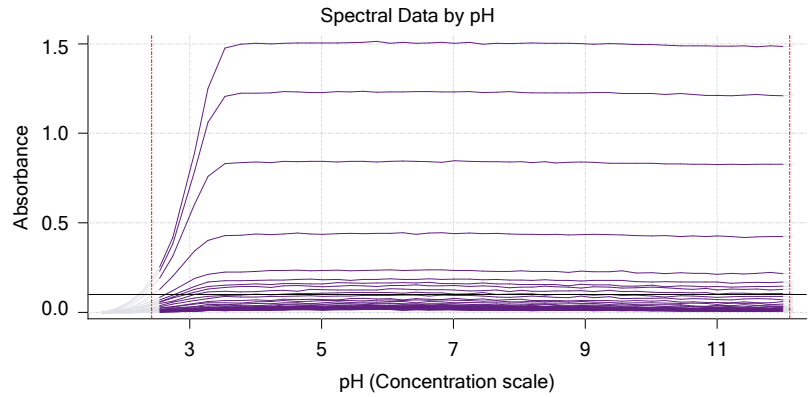



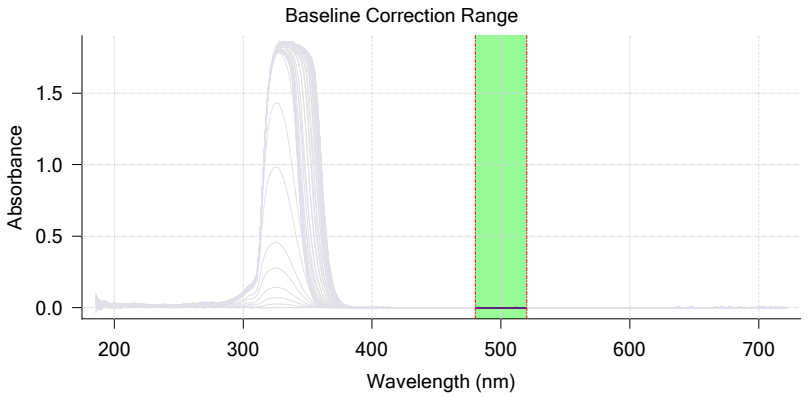



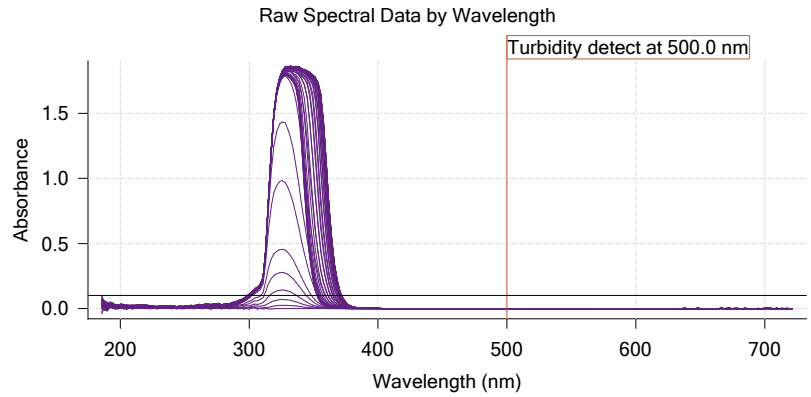
Sample name: **pyridoxine HCl**
Assay name: **pH-metric pKa**
Assay ID: **17I-11015**
Filename: **C:\Sirius_T3\Mehtap\20170911_test_comp_pKa\17I-11015_pyridoxine HCl_pH-metric pKa.t3r**

Experiment start time: **9/11/2017 4:00:50 PM**
Analyst: **Dorothy Levorse**
Instrument ID: **T311053**

## Other graphs (continued)




## Assay Model

| Settings | Value | Date/Time changed | Imported from |
|---|---|---|---|
| Sample name | pyridoxine HCl | 9/11/2017 2:25:57 PM | User entered value |
| Sample by | Weight | | Default value |
| Sample weight | 0.003220 g | 9/11/2017 2:51:35 PM | User entered value |
| Formula weight | 205.64 g/mol | 9/11/2017 2:25:57 PM | User entered value |
| Solubility | Unknown | | Default value |
| Molecular weight | 169.18 | 9/12/2017 11:33:17 AM | User entered value |
| Individual pKa ionic environments | No | | Default value |
| Number of pKas | 2 | 9/12/2017 11:40:37 AM | User entered value |
| Sample is a | Ampholyte | 9/12/2017 11:35:04 AM | User entered value |
| pKa 1 | 2.69 | 9/12/2017 11:13:47 AM | C:\Sirius_T3\Mehtap-files\20170911_test_compounds_pKa |
| Type | Base | | Default value |
| pKa 2 | 4.83 | 9/12/2017 11:35:02 AM | User entered value |
| Type | Acid | 9/12/2017 11:35:14 AM | User entered value |
| logp (XH2 +) | -10.00 | | Default value |
| logP (neutral XH) | -10.00 | | Default value |
| logP (X -) | -10.00 | 9/11/2017 2:25:57 PM | User entered value |
| Stoichiometry | 1.00000 | | Default value |
| Aprotic counterion name | Chloride | | From standards.xml file |
| Stoichiometry | 1.00 | 9/12/2017 11:30:18 AM | User entered value |
| Charge per counterion | -1 | | From standards.xml file |

## Assay Settings

| Setting | Value | Original Value | Date/Time changed | Imported from |
|---|---|---|---|---|
| ***General Settings*** | | | | |
| Analyst name | Dorothy Levorse | | | |
| ***Standard Experiment Settings*** | | | | |
| Number of titrations | 1 | | | |

Sample name: **pyridoxine HCl**
Assay name: **pH-metric pKa**
Assay ID: **17I-11015**
Filename: **C:\Sirius_T3\Mehtap\20170911_test_comp_pKa\17I-11015_pyridoxine HCl_pH-metric pKa.t3r**

Experiment start time: **9/11/2017 4:00:50 PM**
Analyst: **Dorothy Levorse**
Instrument ID: **T311053**

## Assay Settings (continued)

| Setting | Value | Original Value | Date/Time changed | Imported from |
|---|---|---|---|---|
| Minimum pH | 1.800 | | | |
| Maximum pH | 12.200 | | | |
| pH step between points of | 0.200 | | | |
| Minimum titrant addition | 0.00002 mL | | | |
| Maximum titrant addition | 0.10000 mL | | | |
| Argon flow rate | 100% | | | |
| ***Advanced General Settings*** | | | | |
| Detect turbidity using | Spectrometer | | | |
| Monitor at a wavelength of | 500.0 nm | | | |
| Absorbance threshold of | 0.100 | | | |
| Collect turbidity sensor data | No | | | |
| Stir after titrant addition for | 5 seconds | | | |
| For titrant addition, stir at | 15% | | | |
| Start titration using | Cautious pH adjust | | | |
| ***Titrant Pre-Dose*** | | | | |
| Titrant pre-dose | None | | | |
| ***Assay Medium*** | | | | |
| Cosolvent in use | No | | | |
| ISA water volume | 1.50 mL | | | |
| Water added | Automatic | | | |
| After water addition, stir for | 5 seconds | | | |
| At a speed of | 10% | | | |
| ***Sample Sonication*** | | | | |
| Sonicate | No | | | |
| ***Sample Dissolution*** | | | | |
| Perform a dissolution stage | Yes | | | |
| Adjust and hold pH for dissolution | To start pH | | | |
| Stir to dissolve for | 120 seconds | | | |
| For dissolution, stir at | 10% | | | |
| ***Carbonate purge*** | | | | |
| Perform a carbonate purge | No | | | |
| ***Temperature Control*** | | | | |
| Wait for temperature | Yes | | | |
| Required start temperature | 25.0°C | | | |
| Acceptable deviation | 0.5°C | | | |
| Time to wait | 60 seconds | | | |
| Stir speed of | 10% | | | |
| ***Titration 1*** | | | | |
| Titrate from | Low to high pH | | | |
| Adjust to start pH | Yes | | | |
| After pH adjust stir for | 10 seconds | | | |
| ***Data Point Stability*** | | | | |
| Stir during data point collection | Yes | | | |
| For point collection, stir at | 15% | | | |
| Delay before data point collection | 0 seconds | | | |
| Number of points to average | 20 points | | | |
| Time interval between points | 0.50 seconds | | | |
| Required maximum standard deviation | 0.00500 dpH/dt | | | |
| Stability timeout after | 60 seconds | | | |
| ***Experiment cleanup*** | | | | |
| Adjust pH to cleanup | To start pH | | | |
| And then stir for | 60 seconds | | | |
| For cleaning, stir at | 25% | | | |
| Then add water volume | 0.25 mL | | | |
| And then stir for | 30 seconds | | | |

Sample name: **pyridoxine HCl**
Assay name: **pH-metric pKa**
Assay ID: **17I-11015**
Filename: **C:\Sirius_T3\Mehtap\20170911_test_comp_pKa\17I-11015_pyridoxine HCl_pH-metric pKa.t3r**
Experiment start time: **9/11/2017 4:00:50 PM**
Analyst: **Dorothy Levorse**
Instrument ID: **T311053**

## Calibration Settings

| Setting | Value | Date/Time changed | Imported from |
|---|---|---|---|
| Four-Plus alpha | 0.079 | 9/11/2017 4:00:50 PM | C:\Sirius_T3\HCl17I11.t3r |
| Four-Plus S | 1.0031 | 9/11/2017 4:00:50 PM | C:\Sirius_T3\HCl17I11.t3r |
| Four-Plus jH | 1.1 | 9/11/2017 4:00:50 PM | C:\Sirius_T3\HCl17I11.t3r |
| Four-Plus jOH | -0.5 | 9/11/2017 4:00:50 PM | C:\Sirius_T3\HCl17I11.t3r |
| Base concentration factor | 1.015 | 9/12/2017 11:13:47 AM | C:\Sirius_T3\Mehtap-files\20170911_test_compounds_pKa\KOH17I11.t3r |
| Acid concentration factor | 1.002 | 9/11/2017 4:00:50 PM | C:\Sirius_T3\HCl17I11.t3r |

## Instrument Settings

| Setting | Value | Batch Id | Install date |
|---|---|---|---|
| Instrument owner | Merck | | |
| Instrument ID | T311053 | | |
| Instrument type | T3 Simulator | | |
| Software version | 1.1.3.0 | | |
| Dispenser module | | T3DM1100253 | 3/31/2009 6:24:52 AM |
| Dispenser 0 | Water | | 3/31/2009 6:25:05 AM |
| Syringe volume | 2.5 mL | | |
| Firmware version | 1.2.1(r2) | | |
| Titrant | Water (0.15 M KCl) | 8-18-17 | 9/8/2017 9:22:43 AM |
| Dispenser 2 | Acid | | 3/31/2009 6:25:11 AM |
| Syringe volume | 0.5 mL | | |
| Firmware version | 1.2.1(r2) | | |
| Titrant | Acid (0.5 M HCl) | 166940 | 9/8/2017 9:21:27 AM |
| Dispenser 1 | Base | | 3/31/2009 6:25:21 AM |
| Syringe volume | 0.5 mL | | |
| Firmware version | 1.2.1(r2) | | |
| Titrant | Base (0.5 M KOH) | 01/06/17 | 9/8/2017 9:20:03 AM |
| Dispenser 5 | Cosolvent | | 3/31/2009 6:26:24 AM |
| Syringe volume | 2.5 mL | | |
| Firmware version | 1.2.1(r2) | | |
| Distribution valve 5 | Distribution Valve | | 3/31/2009 6:28:19 AM |
| Firmware version | 1.1.3 | | |
| Port A | Methanol (80%, 0.15 M KCl) | 8-15-17 | 9/7/2017 3:40:52 PM |
| Dispenser 3 | Buffer | | 8/3/2010 6:05:16 AM |
| Syringe volume | 0.5 mL | | |
| Firmware version | 1.2.1(r2) | | |
| Titrant | Phosphate Buffer | | 8/15/2017 11:15:20 AM |
| Dispenser 6 | Octanol | | 10/22/2010 11:52:43 AM |
| Syringe volume | 0.5 mL | | |
| Firmware version | 1.2.1(r2) | | |
| Titrant | Octanol | | 8/15/2017 11:16:41 AM |
| Titrator | | T3TM1100153 | 3/31/2009 6:24:17 AM |
| Horizontal axis firmware version | 1.17 AI1DI2DO2 Stepper 2 | | |
| Vertical axis firmware version | 1.17 AI1DI2DO2 Stepper 2 | | |
| Chassis I/O firmware version | 1.11 AI1DI0DO4 Norgren I/O | | |
| Probe I/O firmware version | 1.1.1 | | |
| Electrode | T3 Electrode | T3E0769 | 8/15/2017 10:21:54 AM |
| E0 calibration | -4.08 mV | | 9/11/2017 4:01:14 PM |
| Filling solution | 3M KCl | KCL095 | 9/10/2017 10:38:19 AM |
| Liquids | | | |
| Wash 1 | 50% IPA:50% Water | | 9/11/2017 9:22:00 AM |
| Wash 2 | 0.5% Trition X-100 in H20 | | 9/11/2017 9:22:03 AM |
| Buffer position 1 | pH7 Wash | | 9/11/2017 9:22:06 AM |
| Buffer position 2 | pH 7 | | 9/11/2017 9:22:10 AM |
| Storage position | | | 9/11/2017 9:23:34 AM |
| Wash water | 2.4e+003 mL | 8-16-17 | 8/16/2017 4:10:31 PM |
| Waste | 9.2e+003 mL | | 8/15/2017 10:25:02 AM |
| Temperature controller | | | 8/5/2010 7:35:13 AM |

Sample name: **pyridoxine HCl**
Assay name: **pH-metric pKa**
Assay ID: **17I-11015**
Filename: **C:\Sirius_T3\Mehtap\20170911_test_comp_pKa\17I-11015_pyridoxine HCl_pH-metric pKa.t3r**

Experiment start time: **9/11/2017 4:00:50 PM**
Analyst: **Dorothy Levorse**
Instrument ID: **T311053**

## Instrument Settings (continued)

| Setting | Value | Batch Id | Install date |
|---|---|---|---|
| Turbidity detector | | | 3/31/2009 6:24:45 AM |
| Spectrometer | | 072390 | 11/23/2010 12:22:28 PM |
| Dip probe | | 11086 | |
| Wavelength coefficient A0 | 185.563 | | |
| Wavelength coefficient A1 | 2.17439 | | |
| Wavelength coefficient A2 | -0.000285622 | | |
| Total lamp lit time | 65:29:35 | | 11/23/2010 12:22:28 PM |
| Calibrated on | 9/6/2017 9:33:02 AM | | |
| Integration time | 11 | | |
| Scans averaged | 10 | | |
| Autoloader | | T3AL1100237 | 11/10/2015 10:34:13 AM |
| Left-right axis firmware version | 1.17 AI1DI2DO2 Stepper 2 | | |
| Front-back axis firmware version | 1.17 AI1DI2DO2 Stepper 2 | | |
| Vertical axis firmware version | 1.17 AI1DI2DO2 Stepper 2 | | |
| Chassis I/O firmware version | 1.11 AI1DI0DO4 Norgren I/O | | |
| Configuration | | | |
| Alternate titration position | Titration position | | |
| Alternate reference position | Reference position | | |
| Maximum standard vial volume | 3.50 mL | | |
| Maximum alternate vial volume | 25.00 mL | | |
| Automatic action idle period | 5 minute(s) | | |
| Titrant tube volume | 1.3 mL | | |
| Syringe flush count | 3.50 | | |
| Flowing wash pump volume | 20.0 mL | | |
| Flowing wash stir duration | 5 s | | |
| Flowing wash stir speed | 30% | | |
| Solvent wash stir duration | 5 s | | |
| Solvent wash stir speed | 30% | | |
| Surfactant wash stir duration | 5 s | | |
| Surfactant wash stir speed | 30% | | |
| E0 calibration minimum number of points | 10 | | |
| E0 calibration maximum standard deviation | 0.01500 | | |
| E0 calibration timeout period | 60 s | | |
| E0 calibration stir duration | 5 s | | |
| E0 calibration preparation stir speed | 30% | | |
| E0 calibration buffer wash stir duration | 5 s | | |
| E0 calibration buffer wash stir speed | 30% | | |
| E0 calibration reading stir speed | 0% | | |
| Spectrometer calibration stir duration | 5 s | | |
| Spectrometer calibration stir speed | 30% | | |
| Spectrometer calibration wash pump volume | 20.0 mL | | |
| Spectrometer calibration wash stir duration | 5 s | | |
| Spectrometer calibration wash stir speed | 30% | | |
| Overhead dispense height | 10000 | | |

## Refinement Settings

| Setting | Value | Default value |
|---|---|---|
| Turbidity detection method | Spectrometer | Spectrometer |
| Turbidity wavelength to assess | 500.0 nm | 500.0 nm |
| Turbidity maximum absorbance | 0.100 | 0.100 |
| Turbidity probe threshold | 50.00 | 50.00 |